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Flow-Through Bioassay for Measuring Bioaccumulation of Toxic Substances from Sediment

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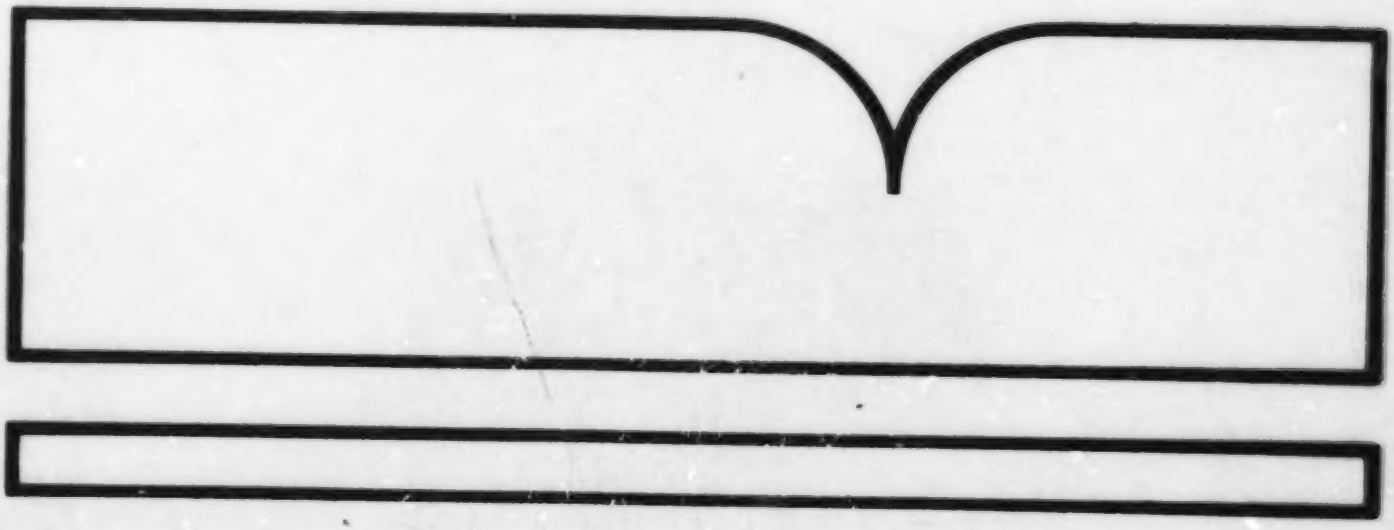

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16. ABSTRACT

A bioassay was developed for testing the solid phase of dredged material that measures the survival of organisms, and the bioaccumulation of toxic substances. This bioassay is intended to estimate the bioaccumulation potential from sediments that are not acutely toxic to test organisms, but are suspected of containing persistent contaminants. Two species of fish, Pimephales promelas and Perca flavescens, and two invertebrate species, Octalasion tyrtaeum and Corbicula fluminea were used in the evaluation of this flow-through bioassay. Although not intended as a toxicity test per se, the test enables interpretation of any mortality that may occur during bioassays.

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FOREWORD

The Great Lakes National Program Office (GLNPO) of the United States Environmental Protection Agency was established in Region V, Chicago, to focus attention on the significant and complex natural resource represented by the Great Lakes.

GLNPO implements a multi-media environmental management program drawing on a wide range of expertise represented by universities, private firms, State, Federal, and Canadian governmental agencies, and the International Joint Commission. The goal of the GLNPO program is to develop programs, practices and technology necessary for a better understanding of the Great Lakes Basin ecosystem and to eliminate or reduce to the maximum extent practicable the discharge of pollutants into the Great Lakes system. GLNPO also coordinates U.S. actions in fulfillment of the Great Lakes Water Quality Agreement of 1978 between Canada and the United States of America.

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Introduction

Over 10 million cubic meters of sediment are dredged annually from Great Lakes waterways. Because much of this material is taken from harbors, connecting channels, and other nearshore areas that often are contaminated with toxic substances, the sediments proposed for dredging need to be evaluated for the presence of bioavailable contaminants and the potential for toxicity to the biota. Sound decisions on the appropriate disposal of the dredged material can be made only after such an evaluation. Presently, no standardized procedure exists for evaluating dredged material in freshwater systems although criteria for discharge of dredged material into marine waters have been developed (USEPA/CE 1977). In the ocean discharge guidelines, it is recommended that bioassays be conducted on liquid, solid, and suspended particulate phases of dredged material. Because it appears that the solid phase has the greatest potential for environmental damage and because measurements of bioaccumulation must be made to evaluate sediments for disposal (USEPA/CE 1977, Seelye and Mac 1983), we developed a bioassay for testing the solid phase of dredged material that measures the survival of organisms and, perhaps more important, the bioaccumulation of toxic substances. Although other workers have demonstrated the bioaccumulation of toxic substances by aquatic organisms from naturally contaminated sediments (Peddicord et al. 1980; Rubinstein et al. 1980, 1983; Seelye et al. 1982), several have used testing methods that result in unacceptable mortality to control organisms (Bahnick et al. 1981, Prater et al. 1983).

Our bioassay is intended to estimate the potential for bioaccumulation of contaminants from sediments that are not acutely toxic to test organisms, but

are suspected of containing persistent contaminants. By using test organisms that are not highly susceptible to toxic compounds, the bioaccumulation test allows estimation of the potential food-chain accumulation of contaminants that may occur in local biota from surficial sediments. In practice, bioaccumulation observed in this bioassay by organisms exposed to test sediments (sediments to be dredged) would be compared to bioaccumulation observed from sediments collected from a reference site (e.g. a disposal site or open lake), and also from control sediments (relatively clean sediment). Decisions could then be based on a comparison of results between test and reference sediments to determine if disposal would cause degradation to the habitat, and between reference and control sediment to determine if even the reference material is seriously contaminated. Although the test is not intended to be a toxicity test per se, use of test, reference, and control sediments enables interpretation of any mortality of organisms that may occur during the bioassays. High mortality in bioassays with test or reference sediment would indicate acute toxicity of sediments in the project area. However if high mortality occurs in all three sediments, it can be assumed that the organisms were not in a healthy state at the time of testing.

We describe the results of 10-day sediment bioassays in which both mortality and bioaccumulation were measured in four aquatic organisms. We exposed two infaunal organisms and two species of fish to test and control sediments in the laboratory.

Materials and Methods

Sediment bioassays were conducted in a flow-through system consisting of eight 39-L glass tanks (Fig. 1). Each tank received 100 mL/min of 20°C water,

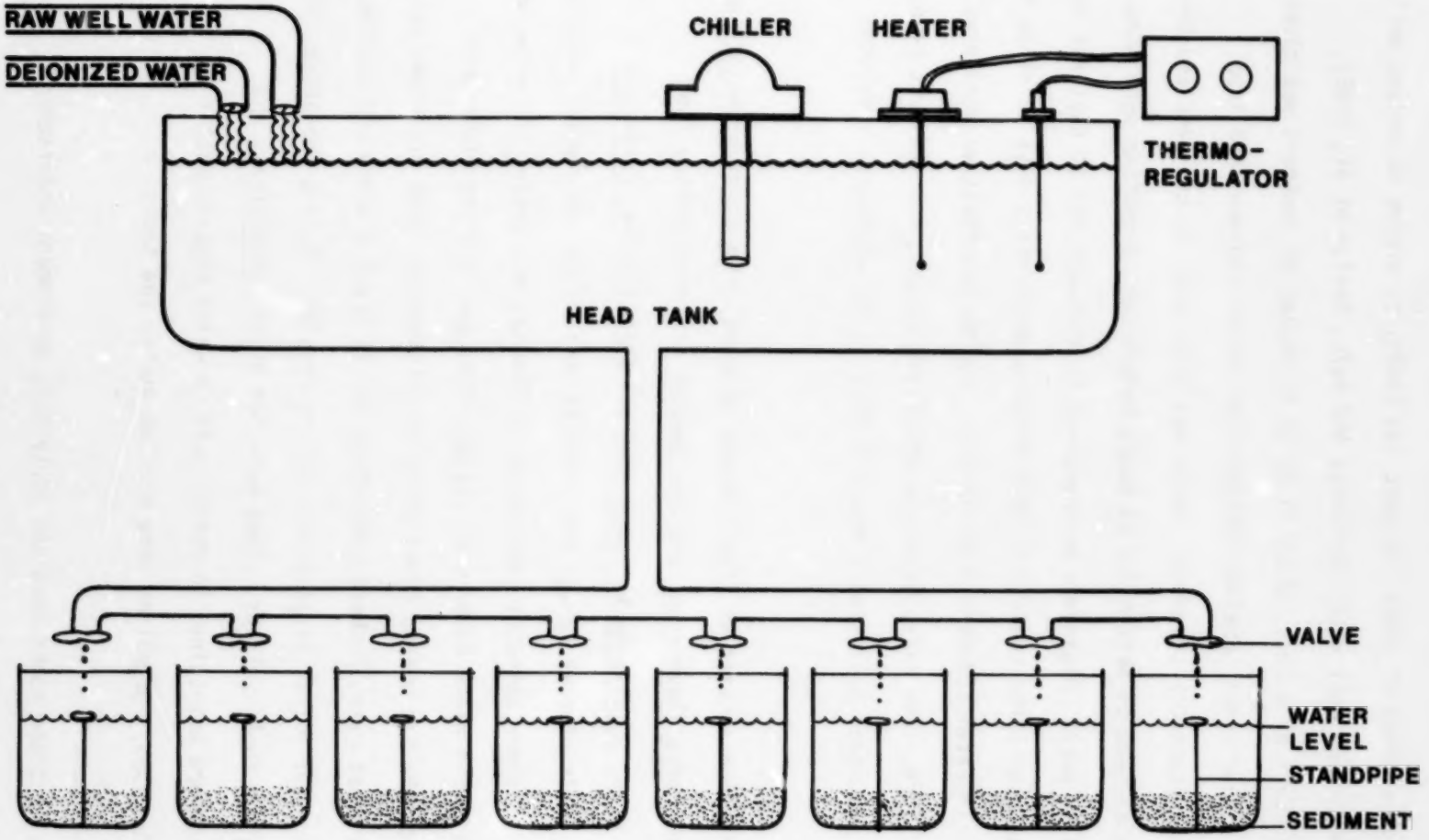

Figure 1. Diagram of sediment exposure system.

softened to a hardness of about 120 mg/L (as $CaCO_3$) by mixing deionized well water with processed well water (hardness 442 mg/L, Seelye et al. 1982). Prior to the start of a test, about 11 kg (5 cm depth) of sediment was added to each tank; four tanks received contaminated (test) sediment and four received clean (control) sediment. Water was then added to the tank and the sediment was allowed to settle for 24 hours before test organisms were added. Tests lasted 12 days. Organisms were exposed to sediment for 10 days and then moved to identical tanks containing only flowing water for 2 days to allow for clearance of ingested sediment from the gut. During the tests we monitored water temperature, flow rate, hardness, dissolved oxygen, and sediment redox potential. Suspended solids were measured only in the exposures involving fish.

We tested two species of fish (fathead minnows, Pimephales promelas, and yellow perch, Perca flavescens) and two species of invertebrates (an oligochaete worm, Octolasion tyrtaeum, and the Asiatic clam, Corbicula fluminea). Adult minnows (2-3g) and juvenile perch (2g) were obtained from the National Fishery Research Laboratory, La Crosse, Wisconsin; the worms were collected from the Black River near Onaway, Michigan; and the clams were obtained from the Sacramento River delta in California. Test organisms were held at the Great Lakes Fishery Laboratory for at least 2 weeks and acclimated to softened water for at least 5 days before testing. During this time, fish were fed Silver Cup[1/] pellets, clams were fed algae (Chlorella), and oligochaetes were maintained in forest duff in which organic matter was available for food. Organisms were not fed during the tests.

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During testing, we also examined the suitability of two other invertebrates: Chironomus larvae and Hexagenia limbata. Chironomus larvae are easily cultured and an important food chain member in the Great Lakes. We rejected them as a test organism, however, because their small size would require large numbers of similarly aged individuals to obtain enough tissue for contaminant analysis. Hexagenia limbata is another important food item in the Great Lakes. We observed high mortality during both holding and testing of this species. High mortality of H. limbata has occurred in other published sediment studies (Bahnick et al. 1981). Because of the small size of Chironomus and the high mortality of H. limbata, no further tests of these organisms were attempted.

Either 10 fish, 14 oligochaete worms, or 30 clams were placed into each tank at the start of the bioassay. We netted fish and collected worms and clams by hand following 10 days of exposure to sediments. At the end of a test, we froze all live organisms whole for contaminant analysis. In addition, a sample of test organisms was frozen prior to the start of the test for determination of preexposure contaminant concentrations. All organisms were thawed and the clams shelled before homogenization and analysis.

Control and test sediments were collected just before each test to minimize chemical changes in the sediment caused by storage (Table 1). Three sets of test sediments were collected with a ponar dredge from the Raisin River near Monroe, Michigan (41° 54' 1" N, 83° 21' 18" W), and three sets of control sediments were shoveled from Meadowood Pond in Saline Township, Michigan (42° 7' 44" N, 83° 47' 45" W). In the first exposure, two sets of tanks were used with fathead minnows in one set and oligochaetes in the other. Yellow perch were tested in sediments from the second collection and clams

Table 1. Physical and chemical characteristics of sediments from the three collections. Dates of collection and dates of various tests are also presented.

Exposures and dates of test	Date Collected	Physical composition (% dry wt.)				Contaminants (ug/g dry wt.)	
		Sand	Silt	Clay	Volatile solids	PCBs	Zn
1. Fathead minnows and oligochaetes (10/20-11/2)							
Control	10/12	65	19	13	2	0.016	34
Test	9/30	11	43	35	8	31.72	244
2. Yellow perch (11/16-11/28)							
Control	11/9	51	24	22	3	0.013	31
Test	10/25	67	16	11	4	12.78	147
3. Asiatic clams (1/28-2/9)							
Control	1/13	16	41	32	10	0.014	67
Test	1/14	36	33	22	7	19.60	162

were tested in sediments from the third collection. Subsamples of sediment were taken before placement in test tanks and frozen for later chemical and physical analysis.

Sediments and test organisms were analyzed for PCBs and Zn to examine the bioaccumulation potential of both an organic and an inorganic contaminant directly from contaminated sediment. We analyzed PCBs in sediment and in test organisms by gas chromatography (GC), using methods previously described by Seelye et al. (1982). Samples of tissue and sediment for Zn analysis were digested in HNO_3 according to the method of May and Brumbaugh (1982) except that no perchloric acid was used, and Zn concentrations were determined on a Perkin Elmer Model 228 atomic absorption spectrophotometer equipped with an HGA2200 graphite furnace. Absorption values for 50-uL samples were compared at 307.6 nm with known standards. Graphite furnace conditions were as follows: nitrogen flow, 20 mL/min; sample drying time, 60 sec with a 15-sec ramp (20-120°C); ashing time, 50 sec with a 10-sec ramp (120° -500°C); and atomization, 6 seconds at 2200°C.

We report concentrations of PCBs and Zn in both sediment and tissue on a dry weight basis to alleviate discrepancies caused by varying water content. Thus subsamples of all analytical samples were dried to measure water content. A wet-to-dry conversion factor was calculated and applied to measured wet-weight concentrations.

We compared PCB and Zn concentrations in test organisms between preexposure samples and test and control samples after exposure, using analysis of variance (ANOVA). For both species of fish and the oligochaetes, all surviving test organisms in a tank were composited to form one analytical

sample. Thus different tanks were considered replicates. Clams remaining alive at the completion of the bioassay were divided into two analytical samples so that within- and between-tank replicates, as well as replicate tanks, were considered in the ANOVA.

Results

The two sediments used provided contrasting levels of PCBs and Zn. Concentrations in test sediment (micrograms per gram, dry weight) ranged from 12.8 to 31.7 PCB and from 147 to 241 for Zn whereas those in control sediment were < 0.02 for PCB and from 31 to 67 for Zn (Table 1). Although sediment was collected three times, which resulted in some variation in both physical and chemical composition, a large difference in the two contaminants of interest between control and test sediment was always found.

Use of the flow-through bioassay produced nearly constant conditions throughout testing, and mortality was low ($\leq$ 8.3%) in test organisms, indicating that the test sediments were not acutely toxic to the organisms tested (Table 2). Mortality was high in only one test where a mechanical failure restricted flow to two tanks holding oligochaetes during exposure. In these two tanks, all oligochaetes died and were not analyzed. Thus in the contaminated sediment treatment only two replicates are reported.

All test organisms accumulated significant ($P \leq 0.05$) amounts of PCBs from test sediments when compared with either organisms exposed to control sediments or preexposure organisms (Table 3). Bioaccumulation factors (BF = dry wt concentration in tissue divided by dry wt concentration in sediment) for organisms exposed to test sediments indicated that oligochaetes were the most

Table 2. Summary of test conditions and mortality of organisms.

Organism and exposure (No. of organisms in parentheses)	Test Conditions				Mortality (%)
	Temp. (°C)	Dissolved oxygen (mg/L)	Hardness (mg/L, $CaCO_3$)	Sediment Eh (mV)	
Fathead minnows					
Control (10)	20.0	7.8	158.6	-146.3	5.0
Test (10)	21.2	7.5	158.6	-175.9	0.0
Perch					
Control (10)	20.2	7.9	128.1	-227.5	0.0
Test (10)	20.2	7.7	128.7	-219.7	0.0
Oligochaetes					
Control (14)	21.2	8.0	150.4	-154.5	5.4
Test (14)	21.0	7.9	156.0	-214.3	0.0
Clams					
Control (30)	19.9	8.5	113.8	-272.5	8.3
Test (30)	19.4	8.4	113.3	-271.5	8.3

Table 3. Mean weight, lipid content, and PCB and Zn concentrations in analyzed samples. Standard errors in parentheses. All sample sizes equal 4 except clams (N=8) and oligochaetes (N=2).

Organism and exposure	Weight (g)	Lipid (%)	Contaminants (ug.g dry wt.) PCB	Zn
Fathead minnows				
Preexposure	2.42 (0.16)	8.5 (0.29)	1.0 (0.02)	189.8 (13.36)
Control	2.08 (0.08)	8.1 (0.18)	1.4 (0.04)	227.6* (13.34)
Test	2.06 (0.09)	8.1 (0.08)	45.4* (1.96)	179.2 (3.95)
Yellow perch				
Preexposure	2.10 (0.04)	4.9* (0.27)	1.6 (0.0)	113.5 (6.92)
Control	1.90 (0.03)	3.9 (0.29)	2.0 (0.18)	128.6 (8.15)
Test	1.98 (0.09)	3.6 (0.23)	8.9* (0.73)	118.4 (12.79)
Oligochaetes				
Preexposure	0.94* (0.07)	0.6 (0.07)	0.4 (0.06)	182.9 (24.96)
Control	0.64 (0.04)	0.6 (0.12)	0.5 (0.08)	141.0 (11.47)
Test	0.58 (0.02)	0.5 (0.0)	125.5* (1.88)	171.0 (36.64)
Asiatic clams				
Preexposure	1.05 (0.03)	1.5 (0.23)	0.8 (0.17)	135.1 (9.64)
Control	1.22 (0.04)	1.8 (0.04)	1.1 (0.04)	97.1* (2.55)
Test	1.12 (0.05)	1.9 (0.10)	3.4* (0.14)	117.8 (3.18)

*Denotes significant difference ($P \leq 0.05$) from other two treatments based on analysis of variance and Duncan's multiple range test.

efficient accumulators (BF, 4) and clams were the least efficient (BF, of 0.2). Both species of fish accumulated PCB--the BF was ' 4 in fathead minnows and 0.7 in yellow perch.

None of the organisms exposed to test sediments accumulated Zn in any of the bioassays, although we did observe several statistically significant changes in the Zn concentration in organisms from other treatments. Clams exposed to control sediment were significantly lower ($P < 0.001$) in Zn concentration than clams before exposure or those held in test sediments (Table 3). Apparently clams had high levels of Zn in tissues when we received them and some depuration occurred in clean sediments. Fatheads exposed to control sediments had significantly higher ($P = 0.03$) concentrations of Zn than fatheads before exposure or those exposed to test sediments (Table 3). This observation is unexplained.

The weight of oligochaetes and the lipid content of yellow perch both decreased significantly during the bioassay (Table 3). The general trend of decreasing weight and lipid content in both species of fish and in the oligochaetes was expected because the organisms were not fed during the 12-day test.

Discussion

System performance

The flow-through bioassay that we evaluated for use in assessing the potential of bioaccumulation from sediments provided conditions allowing for high survival of test organisms. Mor' ty did not exceed 8.3% in species and half the tests resulted in no mortality. The water flow of 100 mL/min was

sufficient for maintenance of good temperature control and a high oxygen saturation (95%).

The 10-day exposure period, as suggested for evaluating dredged material for ocean discharge (USEPA/CE 1977), was seemingly adequate to assess bioaccumulation potential for organic contaminants with similar chemical characteristics as PCBs. However, it is unlikely that test organisms had reached a steady-state concentration , thus the maximum BFs had probably not been attained (Rubinstein et al. 1983). We did not observe the accumulation of Zn from test sediments in this study--perhaps because the Zn in our test sediments was in a form that is biologically unavailable (Engel et al. 1981); increasing the exposure time would not likely have affected the bioaccumulation of Zn. Rubinstein et al. (1983). showed that increasing the time that marine invertebrates were exposed to contaminated sediments from 10 to 100 days did not result in accumulation of Hg or Cd. Seelye et al. (1982) provided evidence that 10 days is sufficient for measuring bioaccumulation potential of biologically available metals from sediments; they reported accumulation of Zn and several other metals by yellow perch in a 10-day exposure. It thus appears that 10 days is sufficient to measure bioaccumulation potential of metals that are in available form; however not all factors that influence this availability are understood.

The effects of not feeding organisms during the test are not certain. It might be argued that withholding food might result in a loss of PCBs as lipids are mobilized and lipophilic contaminants metabolized. However, if this was occurring to any great extent then organisms exposed to control sediments would have lower PCB levels than unexposed organisms. This loss of PCBs was not

observed. Nevertheless, it is possible that feeding would have enhanced accumulation and perhaps feeding of organisms during the test should be attempted. It may not be successful in tests with fish however due to the high turbidity in aquaria caused by resuspension of fine sediments.

Species evaluation

We compared accumulation in two species of fish that have different advantages for use as bioassay test organisms. The yellow perch is widely distributed in the Great Lakes and the young are often found in areas of dredging activity (Barnes 1979). Its commercial and sport fishing value make it an economically important species. On the other hand, the fathead minnow, although also widely distributed in the Great Lakes watershed, is neither as abundant nor as economically important as the perch. It does offer certain other advantages (a) it is routinely available because it is easy to culture, (b) it is tolerant of a wider range of water temperature and dissolved oxygen (Eddy and Underhill 1974), and c) it is widely used as a bioassay organism (Committee on Methods for Toxicity Tests with Aquatic Organism 1975).

Our results show that fathead minnows accumulated higher concentrations of PCBs than did perch. Although this difference was not critical in testing sediments containing high levels of PCBs, the fathead minnow would be the superior test organism for testing sediments with low concentrations of contaminants. Higher lipid levels in fathead minnows (Table 3), as well as behavioral differences between the two species, could account for their greater uptake of PCBs. Fathead minnows were more active at the water sediment interface than were yellow perch, resulting in a greater resuspension of the sediment. This observation is supported by measurements of suspended solids in

tanks of the two species during the bioassay. In control and test sediments, used with fathead minnows suspended solids in control and test sediments were 125.8 and 200.8 mg/L, respectively). In the yellow perch exposure these values were only 23.5 and 27 mg/L.

O. tyrtaeum was shown to be the preferred invertebrate test species because it accumulated PCBs to a greater extent than did the clams. Although this species is not readily available and not a common inhabitant of Great Lakes nearshore areas we feel an oligochaete worm would provide a benthic invertebrate for freshwater testing similar to the polychaete Nereis used in marine sediment evaluation (Rubinstein et al. 1983, Elder et al. 1979). Both Nereis and O. tyrtaeum appear to accumulate organic contaminants readily and are of adequate size to provide ample tissue for analysis.

The Asiatic clam is common in several areas of the country and has been found in western Lake Erie (Scott-Wasilk et al. 1983). However, our results suggest that its suitability as a test species for measuring bioaccumulation is questionable due to several factors: (a) it may cease feeding in certain sediments, (b) it had the lowest BCF of all the species we tested, and (c) the presence of the shell causes confusion as to what to use as an analytical sample. Although Asiatic clams accumulate metals in their shells (Clarke et al. 1979), the ecological and toxicological significance of this metal accumulation is unknown. This leads to uncertainty as to whether shells should be included in the contaminant analysis. Both organic and inorganic analyses are often conducted on samples taken from the same preparation, but the inclusion of shells in the organic analysis could result in analytical problems.

References

Bahnick, D. A., W. A. Swenson, T. P. Markee, D. J. Call, C. A. Anderson, and R. T. Morris. Development of bioassay procedures for defining pollution of harbor sediments, Part I. CLSES Contract Publication No. 56. Center for Lake Superior Environmental Studies, University of Wisconsin, Superior, 1981. 186 pp.

Barnes, M. D. Inventory and review of literature data sources pertaining to ichthyological and fisheries research in the nearshore zone of Lake Erie. Center for Lake Erie Area Research Technical Report No. 127. Ohio State University, Columbus, Ohio, 1979. 82 pp.

Clarke, J. H., A. N. Clarke, D. J. Wilson, and J. J. Frauf. On the use of Corbicula fluminea as indicators of heavy metal contamination. In: Proceedings, First International Corbicula Symposium, J. C. Britton, ed. Texas Christian University Research Foundation, Fort Worth, Texas, 1979. pp. 153-163.

Committee on Methods for Toxicity Tests with Aquatic Organisms. Methods for acute toxicity tests with fish, macroinvertebrates and amphibians. EPA-660/3-75-009, U.S. Environmental Protection Agency, Corvallis, Oregon, 1975. 61 pp.

Eddy, S., and J. C. Underhill. Northern Fishes. University of Minnesota Press, Minneapolis, Minnesota, 1974. 414 pp.

Elder, D. L., S. W. Fowler, and G. G. Polikarpov. Remobilization of sediment associated PCBs by the worm Nereis diversicolor. Bull. Environ. Contam. Toxicol. 21:448-452, 1979.

Engel, D. W., W. G. Sunda, and B. A. Fowler. Factors affecting trace metal uptake and toxicity to estuarine organisms. I. Environmental parameters. In Biological monitoring of marine pollutants, F. J. Vernberg, A. Calabrese, F. P. Thurberg, and W. B. Vernberg, eds. Academic Press, New York, 1981. pp. 127-144.

May, T. W., and W. G. Brumbaugh. Matrix modifier and L'vov platform for elimination of matrix interferences in the analysis of fish tissues for lead by graphite furnace atomic absorption spectrometry. Anal. Chem. 54:1032-1037, 1982.

Peddicord, R., H. Tatem, A. Gibson, and S. Pedron. Biological assessment of upper mississippi river sediments. Misc. Pap. EL-80-5, U.S. Army Engineers Waterways Experiment Station, Vicksburg, Mississippi, 1980. 82 pp.

Prater, B. L., R. L. Bennett, P. J. Crerar, and R. A. Laskowski-Hoke. An evaluation and refinement of a 96-hour sediment bioassay procedure. (Draft report) EPA 68-01-6471. U.S. Environmental Protection Agency, Chicago, Illinois, 1983. 90 pp.

Rubinstein, N. I., C. N. Asaro, and C. Sommers. The effects of contaminated sediments on representative estuarine species and developing benthic communities. In: Contaminants and Sediments, Vol. 1, R.A. Baker, ed. Ann Arbor Science, Ann Arbor, Michigan, 1980. pp. 445-461.

Rubinstein, N. I., E. Lores, and N. R. Gregory. Accumulation of PCBs, mercury and cadmium by Nereis virens, Mercenaria mercenaria and Palaemonetes pugio from contaminated harbor sediments. Aquatic Toxicology 3:249-260, 1983.

Scott-Wasilk, J., G. G. Downing, and J. S. Lietzow. Occurrence of the Asiatic clam Corbicula fluminea in the Maumee River and western Lake Erie. J. Great Lakes Res. 9:9-13, 1983.

Seelye, J. G., R. J. Hesselberg, and M. J. Mac. Accumulation by fish of contaminants released from dredged sediments. Environ. Sci. Technol. 16(8):459-464, 1982.

Seelye, J. G., and M. J. Mac. Bioaccumulation of toxic substances associated with dredging and dredged material disposal: A literature review. EPA AD-14-F-1-529-0. U. S. Environmental Protection Agency, Chicago, Illinois, 1983. 45 pp.

USEPA/CE. Ecological evaluation of proposed discharge of dredged material into ocean waters; implementation manual for Section 103 of Public Law 92-532 (Marine Protection, Research, and Sanctuaries Act of 1972). Environmental Effects Laboratory, U.S. Army Engineer Waterways Experiment Station, Vicksburg, Mississippi, 1977 (Second Printing 1978). 19 pp. and 8 appendices.